Genetics of Resistance to Bacterial Wilt in Maize

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AGRICULTURAL EXPERIMENT STATION
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AND MECHANIC ARTS

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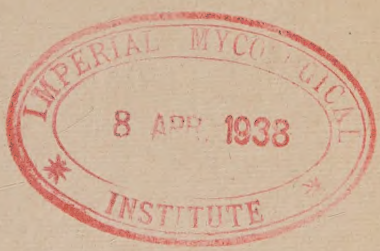

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CONTENTS

Summary 73
Review of literature 76
Materials 77
Pathogen 77
Parental inbreds 77
Methods 78
Experimental data 81
Results of F_1 tests 81
Reaction of progeny from the backcrosses (OSF x WF) x WF and reciprocal, and (OSF x W-134) x W-134 83
Reaction of F_2 progeny from crosses OSF x WF and OSF x W-134 88
Reaction of other crosses 88
Factorial hypothesis for inheritance of resistance 90
Verification of factorial hypothesis by tests of backcross and F_3 families 92
Breeding behavior of backcross families 92
Breeding behavior of F_3 families 99
Linkage relations 104
Cob color and resistance 104
Sugary endosperm and resistance 107
Days to maturity and resistance 108
Ear row number and resistance 109
Discussion and conclusions 111
Literature cited 114

SUMMARY

Highly stable inbred lines were utilized in determining the mode of inheritance of resistance to bacterial wilt in maize caused by *Phytomonas stewarti*. The disease was produced artificially, each plant being inoculated by means of a hypodermic syringe. With the exception of certain tests of F_2 and backcross progeny, a single highly virulent strain of the organism was used throughout.

Many crosses were made between inbreds of various degrees of resistance and tested for dominance relations. In general, the crosses were approximately equal in resistance to that of the more resistant parent involved. A few of the crosses between inbreds intermediate in resistance, however, were more resistant than either of the parents. This was attributed to supplementary action of resistance factors.

Inheritance studies were chiefly confined to the analysis of the later generation progenies of two crosses: OSF, a very resistant inbred of dent corn, x WF, a very susceptible inbred of flint corn, and OSF x W-134, a very susceptible inbred of early yellow sweet corn. Tests of the F_1 progeny showed that dominance of resistance was fairly complete.

Backcross progeny (F_1 x susceptible) of both crosses could readily be divided into four equal groups on the basis of resistance and susceptibility: Very resistant, moderately resistant, susceptible and very susceptible. It was assumed that these different degrees of resistance were due to the independent segregation of two supplementary factors, Sw_1 and Sw_2, completely dominant over their recessive alleles, sw_1 and sw_2, respectively. Accordingly, the very resistant genotypes contain both Sw_1 and Sw_2, moderately resistant genotypes contain only Sw_1, susceptible genotypes possess Sw_2 alone, and the very susceptible genotypes are double recessive, containing neither of the two dominant factors.

In certain of the backcross tests made under different environmental conditions, it was evident that each of the four groups was not a homogeneous lot. Each could be subdivided into two groups, one being slightly higher in resistance than the other. This indicated that a third minor supplementary factor, Sw_3, was involved. According to the data this factor, when alone, produces a degree of resistance only slightly higher than that exhibited by the triple recessive types and when in combination with either or both Sw_1 or Sw_2 modifies their expression by slightly increasing resistance.

Results of F_2 tests and tests of F_3 and backcross families substantiated the above factorial hypothesis. It was concluded, therefore, that at least three (two major and one minor) dominant, independently inherited, supplementary factors are involved in the inheritance of resistance to bacterial wilt in maize. The presence of all three factors either in heterozygous or homozygous dominant conditions ($Sw_1sw_1Sw_2sw_2Sw_3sw_3$ or $Sw_1Sw_1Sw_2Sw_2Sw_3Sw_3$) results in a high degree of resistance, whereas the triple recessive condition ($sw_1sw_1sw_2sw_2sw_3sw_3$) results in a high degree of susceptibility. The various intermediate degrees of resistance or susceptibility between these two extremes may be explained by the presence of only one dominant factor or the different possible combinations of any two.

It was demonstrated that the parental combinations, red cob color and resistance, and white cob color and susceptibility, were more frequent in the backcross and F_2 progeny than the non-parental. This signifies that one of the factors for resistance is genetically linked with the P-gene for cob color. The frequency of the non-parental types indicates that the linkage is not very close.

It was also shown that the parental combinations, late maturity and resistance, and early maturity and susceptibility, tend to remain together in the later generation progenies. This might be attributed to genetic linkage, but it is possible that certain factors for early maturity may also modify resistance.

Endosperm characters such as yellow or white color and starchy or sugary texture seemed to assort independently of resistance.

Genetics of Resistance to Bacterial Wilt in Maize[1]

By E. J. WELLHAUSEN[2]

During 1931-33 bacterial wilt or Stewart's disease of maize, caused by *Phytomonas*[3] *stewarti* (E.F.S.) Bergey *et al* [*Bacterium stewarti* E.F.S.], was more widespread than ever before known. None of the commercial early yellow varieties of sweet corn grown at that time was resistant, and losses often as high as 50 percent in such varieties were encountered. Since resistant strains or varieties offer the only practical means of control for this disease, such losses led to the initiation of an extensive plant breeding program for the development of resistant, early maturing strains of sweet corn. The object of the investigations reported herein was to determine how resistance is inherited. The availability at this station of a large number of different, highly stable inbred lines of maize offered unusual opportunities for investigation of the problem.

At the beginning of the study 56 different inbred lines of maize, including 23 of yellow dent, one of white flint, 18 of white sweet (Evergreen) and 15 of very early sweet corn, mostly yellow, were tested for resistance and susceptibility. In a preliminary paper (12) it was shown that these inbreds may be divided into five classes, ranging from very resistant to very susceptible. In the present paper, data are presented on the mode of inheritance of resistance in crosses between certain highly resistant and very susceptible inbreds. Many crosses between resistant and susceptible lines were made and tested for dominance relations, but inheritance studies were confined chiefly to two crosses: OSF, a very resistant inbred of yellow dent field corn, x WF, a very susceptible inbred of white flint corn, and OSF x W-134, a very susceptible, early, yellow, sweet-corn inbred of Golden Bantam type. Inasmuch as field corn is generally more resistant than sweet corn, an effort was made to determine the genetic relation between resistance and characters which are commonly associated with field corn such as starchy endosperm, red cob, late maturity and general plant vigor.

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[1]Project 404 of the Iowa Agricultural Experiment Station.

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[3]It is probable that neither of the names given above will eventually prove valid. This paper, however, is not one concerned primarily with nomenclature.

REVIEW OF LITERATURE

Many reports on differences in resistance of open-pollinated varieties and inbred strains of corn to bacterial wilt have appeared since the disease was first discovered in 1895, but methods of inheritance of these differences have never been presented.

In general, the literature reveals that the late maturing varieties or strains of corn are more resistant than early maturing ones. One of the more extensive tests of open-pollinated varieties was made by Rand and Cash (7). In 1921 they reported results on 53 varieties of sweet corn and 45 varieties of field corn. It was found that an arrangement of varieties according to maturity coincides very closely with an arrangement according to percentage of wilt. The majority of the dent varieties of field corn were highly resistant, but the early flint varieties invariably were susceptible. The later sweet varieties, such as Zig Zag Evergreen, Stowell's Evergreen and Country Gentleman, consistently gave a low percentage of disease while serious losses occurred in the early varieties of Golden Bantam type.

Similar results in open-pollinated varieties have been reported by Reddy (9), Johnson and Haskell (3), the Ohio Agricultural Experiment Station (6) and others. Practically all the open-pollinated early varieties commonly grown by canners and market gardeners were found to be susceptible.

Attention has turned to the testing of inbred lines and hybrids with the hope that highly resistant strains might be found which could be used in developing early resistant hybrids to replace the wilt-susceptible, popular, early maturing, open-pollinated varieties of sweet corn. Reddy and Holbert (10) in 1928 reported data on the resistance to bacterial wilt of 15 inbred strains of yellow dent corn and 4 hybrids. Reduction in yield of the inbred strains following artificial inoculation ranged from 5.8 to 67.3 percent. First generation crosses appeared to be more resistant than either of the parent strains.

In 1935 Wellhausen (12) reported the results of tests on 56 inbred lines, including dent, flint and sweet corn inbreds. The majority of the dent corn inbreds were resistant; the majority of the inbreds from Evergreen varieties of white sweet corn were intermediate in resistance, and most of the early yellow sweet-corn inbreds and flint inbreds were susceptible. Resistance was found to be dominant in each of the 14 crosses tested. Ivanoff and Riker (2) in 1936 reported studies on resistance of a large number of inbred strains and hybrids tested at De Kalb, Ill. They found a positive correlation between resistance and plant height as well as resistance and late maturity in both inbred strains and hybrids. Resistance in the hybrids was gen-

erally dominant. They also report that hybrid vigor is responsible for some resistance.

Mahoney and Muncie (5), in a report on resistance of inbred strains and hybrids, raised the question whether resistance to bacterial wilt is heritable. Their conclusions indicate that wilt resistance in corn is largely dependent on vigor and rate of growth. In the following pages it will be shown that although earliness and lateness, plant height, vigor and rate of growth may have some effect on resistance, they are not the major heritable factors responsible for resistance or susceptibility.

MATERIALS

PATHOGEN

The culture of bacteria used for resistance tests traces back to a highly virulent strain (S15) obtained from Dr. W. H. Burkholder of Cornell University. This strain was obtained by isolation from diseased sweet corn in New York during the summer of 1932. It proved to be the most virulent among eight other strains tested. A fairly constant degree of virulence was maintained throughout this investigation by frequent, short, host passages and subsequent testing of reisolated colonies on four standard inbred lines of known reaction, including the parental lines used in inheritance studies. In a few cases a strain of S15 which had lost considerable virulence by continued culture on artificial media was used, but, unless otherwise stated, the standardized highly virulent strain was used throughout. The organism were grown on nutrient-dextrose agar at room temperature.

PARENTAL INBREDS

Inbred lines utilized throughout this investigation had been inbred (selfed) for eight or more generations and were highly stable in all characters. The distinguishing features of the parental inbreds involved in the two crosses analyzed (OSF x WF and OSF x W-134) are summarized as follows:

Abbreviation	Description
OSF	Inbred yellow dent, 14-rowed, medium early in maturity, 5 to 6 feet tall when mature, red-cobbed, **very resistant.**
WF	Inbred white flint, 8-rowed, early maturing, 5 to 5½ feet tall when mature, white-cobbed, **very susceptible.**
W-134	Inbred yellow sweet, 8-rowed, very early maturing, 3 to 4 feet tall when mature, white-cobbed, **very susceptible.**

Fig. 1. Reaction of two resistant dent corn inbreds, MCC and OSF, and the susceptible white flint inbred WF in the greenhouse 16 days after inoculation.

Inbred OSF was one of the most resistant dent corn inbreds available at the beginning of this investigation. It readily showed symptoms of the disease shortly after inoculation but soon recovered. After 4 or 5 weeks no signs of the disease, with the exception of a slight degree of stunting, could be detected. Stunting was most apparent in the greenhouse 16 or 20 days after seedling inoculation. At this time inoculated plants were generally from 25-30 percent lower in dry weight than corresponding uninoculated checks. As plants grew older, stunting became less apparent. In the field no stunting was evident from inoculated and uninoculated paired rows at maturity.

Inbreds WF and W-134, in sharp contrast to OSF, were among the most susceptible lines tested. In the greenhouse these lines were completely wilted or dried up within 16 or 20 days after inoculation. In the field WF wilted somewhat more slowly than W-134, but both lines were usually dead within 4 weeks. Figures 1 and 2 show typical reactions of OSF, WF and W-134, together with a few other inbreds of similar resistance or susceptibility under greenhouse conditions. A test of a large number of selfed ears from the eighth and subsequent generations of inbreeding showed that the parental inbreds were homozygous for these specific reactions.

METHODS

The F_1's of the crosses analyzed were selfed and backcrossed to the respective susceptible parent. Representative numbers of the resulting F_2 and backcross progenies were grown in the

field and selfed to produce F_3 and backcross families. The mode of inheritance of resistance was determined from reactions of the F_1, F_2 and backcross progenies to a highly virulent strain of the pathogen. Results obtained from the F_2 and backcross populations were verified by tests of the F_3 and backcross families.

In the greenhouse, plantings were made in flats 16″x22″x3″ (5 rows per flat, 15 seeds per row) and on large benches 5′x27′x6″ in 5-foot rows 6 inches apart and from 45 to 50 seeds per row. Fertile soil was used. Flats and benches were watered amply, and temperatures were maintained around 80° F. except on hot sunny days during late spring and summer. An attempt was made to produce an environment conducive to rapid seedling growth. Such conditions, according to Rand and Cash (8) and E. F. Smith (11), are also conducive to rapid development of the disease.

Fig. 2. Reaction of two early, yellow, sweet-corn inbreds, GB-16 and W-134, in the greenhouse 16 days after inoculation.

Inasmuch as seedlings were tested throughout the year, conditions were necessarily variable due to short cloudy days in winter and extremely high temperatures on hot days in summer. These different environments had a definite effect on expression of symptoms and on manner of segregation obtained in identical genetic material. It was highly important, therefore, that all tests under slightly different environments be analyzed and recorded separately. In the field, plants were grown in rows 3½ feet apart and spaced at about 6-inch intervals within the row.

All plants tested were inoculated in the seedling stage with a standard hypodermic syringe. The syringe was filled with a heavy suspension of bacteria, and a single puncture per seedling was made in the vicinity of the first node with permanent roots (fig. 3). The suspension of bacteria was made in water by washing the organisms from the surface of nutrient-dextrose-agar plates after 18 to 24 hours of growth. A more or less uniform concentration was made up from time to time with the aid of a nepholometer. With this method of inoculation from 98 to 100 percent infection was invariably obtained.

F_1 generations were tested in the same environment with the respective parental inbreds. In these tests, plantings of each, in either greenhouse or field, were made in paired rows one of

Fig. 3. Inoculation of 6-day-old seedlings in the greenhouse.

which was inoculated and the other retained as a check. Notes were taken at weekly intervals on type of symptoms, rate of wilting or death, rate of recovery and degree of stunting which at the end of the test period was often measured on a dry weight basis. The reaction of paired rows of the F_1 in each case was compared with that of the parental inbreds under similar conditions.

The seeds of backcross and F_2 progeny were separated, before planting, on the basis of yellow and white endosperm color in the cross involving the white flint parent, WF, and according to starchy and sugary in the cross involving the sweet corn parent, W-134. Rows of each were then replicated with the respective parental inbreds. Notes were first taken when the majority of the plants of the susceptible parent were dead. At this time the dead plants within the backcross or F_2 populations were counted and removed. The process was repeated at weekly intervals until no more dead plants could be found. Remaining plants were then classified as either healthy or diseased, or doubtful in certain cases, and discarded.

In the greenhouse the F_3 and backcross families were tested on the large benches. Forty-five to fifty seeds of each family were planted in a 5-foot row. With the rows 6 inches apart, 48 families per bench could be tested at one time together with two or three rows of each of the parental inbreds as checks. Families breeding true for colorless or sugary endosperm, as the case may be, were distributed among the others according to their theoretical ratios. In the field 60 seeds of each family were planted in 30-foot rows. Plants in each family were classified on the same basis as that described for the F_2 and backcross populations. Families tested at different times or on different benches were analyzed separately.

EXPERIMENTAL DATA

RESULTS OF F_1 TESTS

Forty-nine crosses were made and tested for resistance. Types of crosses made and number of each type tested are as follows: Two resistant x resistant, 11 resistant x susceptible including reciprocals, 4 resistant x intermediate, 20 intermediate x intermediate and 12 susceptible x susceptible inbreds. Data on these crosses have been presented in detail in a thesis (13) submitted to the graduate faculty of Iowa State College in candidacy for the degree of Doctor of Philosophy. Reaction of the F_1 in most cases was similar to that of the more resistant parent showing fairly complete dominance of resistance. Reciprocal crosses gave

Fig. 4. Reaction of crosses of resistant x susceptible inbreds in comparison to the susceptible inbred WF (16 days after inoculation).

similar results. Typical reactions of very resistant x very susceptible inbreds such as OSF x WF and OSF x W-134 are shown in fig. 4.

Relatively few of the crosses between intermediate lines were thoroughly tested, but certain ones appeared to be markedly more resistant than either of the parents. This probably was not due to hybrid vigor since other combinations of the same apparent vigor did not show marked increases in resistance. Thus the increase in resistance may have been due to complementary or supplementary action of specific factors.

Different combinations of various susceptible inbreds were tested to determine the effect of hybrid vigor and whether certain combinations were more resistant than others of the same apparent vigor. The majority of these crosses was only slightly less susceptible than the respective parents despite marked increases in vigor. Reaction of WF x GB is typical of these crosses under greenhouse conditions (fig. 5). GB was a very susceptible Golden Bantam inbred closely related to W-134. The F_1, in this case, was killed in approximately the same time required to kill the respective parents. Under field conditions all plants of this cross were completely wilted 4 or 5 weeks after inoculation and seldom were over 2 feet in height.

In a few instances, certain combinations having one parent in common showed significant differences in degree of resistance. For example, the F_1 Sto (susceptible yellow dent) x W-134 was more resistant than Sto x GB-797. GB-797 was not quite as susceptible as W-134. In the field the inoculated row of Sto x

GB-797 dried up previous to tasseling, whereas that of Sto x W-134 did not dry up until shortly after the earing stage, permitting the formation of a small ear. Similar differences also were evident in the greenhouse. Check rows in either case showed no difference in hybrid vigor between the two crosses. It is possible, therefore, that the two very susceptible sweet corn inbreds W-134 and GB-797 brought in different minor supplementary factors for resistance.

REACTION OF PROGENY FROM THE BACKCROSSES (OSF x WF) x WF AND RECIPROCAL, AND (OSF x W-134) x W-134

Progeny from the backcross (OSF x WF) x WF were tested with both the standard highly virulent strain and an old culture of S15 which had lost considerable virulence by continued culture on nutrient-dextrose agar. Results of inoculations with the strain of low virulence (test 1) are recorded in table 1, A. For this test and in subsequent tests two classifications are given—early and final. Early or first classification was made when all the plants of the susceptible parent included with each test were dead. Final classification was made when individual plants of the particular population under consideration ceased to die off. In test 1 (table 1, A) 28.5 percent or slightly over one-fourth of the backcross progeny were dead when the first classification was made. Final results, if diseased plants are classed with the dead, closely approximate a 1:1 ratio:

Healthy[4], 885 (905)[5]: *Dead and diseased,* 925 (905), P = .37.

Data obtained with the strain of high virulence (test 2) are given in table 1, B. As in test 1, approximately one-fourth (27.2 percent) of the progeny were dead when the first classification was made, but the final results are in close agreement with a 1:3 ratio:

Healthy, 174 (171) : *Dead and diseased,* 507 (510), P = .74.

According to these data, the backcross progeny may be divided into four groups as follows:

I. Very resistant—Recovered from infection with the highly virulent strain in test 2.

II. Moderately resistant—Attacked by the highly virulent strain in test 2 but, together with the above group, recovered from infection with the strain of low virulence as shown by the 1:1 ratio in test 1.

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[4]The term, Healthy is used to designate those plants which had fully recovered from infection, in so far as all outward signs were concerned.

[5]Theoretical numbers calculated on the basis of the proposed ratio.

TABLE 1. RESULTS OF INOCULATIONS OF BACKCROSS PROGENY FROM CROSSES (OSF x WF) x WF AND RECIPROCAL, AND (OSF x W-134) x W-134.

A. Test No. 1 (Culture of low virulence)

Cross	Pedigree number	Endosperm color or character	No. of plants	Ear'y classification			Final classification				
				Days after inoc.	Alive	Dead	Days after inoc.	Heal.	Doubtful	Dis.	Dead
(OSF x WF) x WF	W-824	Yellow	430	22	311	119	42	210			220
		White	455	22	330	125	42	223			232
	W-826	Yellow	254	22	175	79	42	118		11	125
		White	244	22	174	70	42	121		15	108
	W-827	Yellow	229	22	165	64	42	108		16	105
		White	198	22	138	60	42	105		15	78
		Total	1810		1293	517		885		57	868
		Percent			71.5	28.5		48.8		3.1	47.9
Parents	OSF	Yellow	65	22	62	3	42	62		0	3
	WF	White	122	22	26	96	42	2		0	120

B. Test No. 2 (Culture of high virulence)

(OSF x WF) x WF	W-830	Yellow	134	14	106	28	35	43		29	62
		White	148	14	121	27	35	33		26	89
	W-831	Yellow	200	14	134	66	35	47		20	133
		White	199	14	135	64	35	51		15	133
		Total	681		496	185		174		90	417
		Percent				27.2		25.5		13.2	62.2
Parents	OSF	Yellow	26	14	25	1	35	25		0	1
	WF	White	30	14	0	30					

C. Test No. 3 (Culture of low virulence)

WF x (OSF x WF)	W-816	Yellow	76	18	49	27	42	34			42
		White	80	18	48	32	42	33			47
	W-817	Yellow	123	18	74	49	42	38			85
		White	89	18	57	32	42	41			48
		Total	368		228	140		146			222
		Percent				38.0		39.7			60.3
Parents	OSF	Yellow	15	18	14	1	42	14			1
	WF	White	20	18	5	15	42	0			20

D. Test No. 4 (Culture of high virulence)

WF x (OSF x WF)	W-816	Yellow	208	10	158	50	37	35	23		150
		White	194	10	134	60	37	27	19		148
		Total	402		292	110	37	62	42		298
		Percent				27.3		15.4	10.4		74.1
Parents	OSF	Yellow	12	10	11	1	37	11			1
	WF	White	8	10	1	7	37	1			7

E. Test No. 5 (Culture of high virulence)

F_1 x WF	W-824		200				35	59			141
WF x F_1	W-1183		186				35	44			142

—Continued.

TABLE 1. RESULTS OF INOCULATIONS OF BACKCROSS PROGENY FROM CROSSES (OSF x WF) x WF AND RECIPROCAL, AND (OSF x W-134) x W-134.

F. Test No. 6 (Culture of high virulence)

Cross	Pedigree number	Endosperm color or character	No. of plants	Early classification			Final classification				
				Days after inoc.	Alive	Dead	Days after inoc.	Heal.	Doubtful	Dis.	Dead
(OSFxW-134) x W-134	W-1131	Starchy	67	20	51	16	58	16			51
		Sugary	65	20	43	22	58	15			50
	W-1132	Starchy	96	20	82	14	58	24			72
		Sugary	73	20	59	14	58	22			51
		Total	301		235	66		77			224
		Percent				22		25.6			74.4
Parents	OSF	Starchy	20	20	20	0	58	20			0
	W-134	Sugary	24	20	2	22	58	0			24

G. Test No. 7 (Culture of high virulence)

(OSFxW-134) x W-134	W-1160	Starchy	379	20	179	200	40	89		29	261
		Sugary	371	20	176	195	40	106		31	234
		Total	750		355	395		195		60	495
		Percent				52.6		26.0		8.0	66.0

III. Susceptible—Killed by either strain of the organism but distinguished in test 1 as the group of plants dying after all plants of the susceptible parent had died.

IV. Very susceptible—The group of plants which reacted like plants of the susceptible parent.

Fig. 5. Reaction of a cross between two susceptible inbreds in the greenhouse 16 days after inoculation. Flat on left is an uninoculated duplicate of one on right.

Such results may be explained by the independent segregation of two dominant supplementary factors, one of which is capable of producing a higher degree of resistance than the other. Upon further consideration of the final data of test 2 (table 1, B), however, it is possible that a third, minor or modifying, factor is involved. When the diseased plants were classed with the dead a 1:3 ratio resulted, but on the basis of three classes—healthy, diseased and dead—a 2:1:5 ratio is revealed:

Healthy, 174 (170) : *Diseased,* 90 (86) : *Dead,* 417 (425), P = .80.

This shows that at least group II may be equally divided into two sub-groups, one of which is slightly more susceptible than the other and suggests that the same may be true of the other groups.

No linkage between resistance and yellow or white endosperm is evident from data in either table 1, A or B. The X^2 test for independence was applied to these data according to methods given by Fisher (1). The value of P was .31 for the final data in table 1, A; and .43 for those in table 1, B.

Test 3 (table 1, C) involves progeny from the reciprocal backcross WF x (OSF x WF). Plants in this test were inoculated with the strain of low virulence, but conditions were more favorable for disease development than in test 1. The percentage of plants dead when first notes were taken is not comparable to that of test 1 and 2. Considerable variation between tests may be expected in the early classifications because very often the plants of the susceptible parent did not die uniformly. In such cases it was purely an arbitrary matter as to when the check should be pronounced dead and first notes taken. Final results of test 3 approximate a 3:5 ratio:

Healthy, 146 (138) : *Dead,* 222 (230), P = .40.

This ratio is half way between a 1:1 and a 1:3 which again shows that group II consists of at least two kinds of genotypes slightly different in resistance.

Test 4 (table 1, D) involves progeny of the same reciprocal backcross. This test was made in July under high temperature conditions which are very conducive to rapid wilting. The highly virulent strain was used. When the majority of the plants of the susceptible parent were dead, 27.3 percent of the test plants were also dead. This agrees with the early classifications in tests 1 and 2. Final results, on the basis of alive and dead, closely approximate a 1:3 ratio:

Alive, 104 (100) : *Dead,* 298 (302), P = .69.

Approximately 40 percent of the plants remaining alive, however, were classified as doubtful. This signifies that the very resistant group I may also consist of two slightly different genotypes. In this test the disease was so much more severe that all the plants of group II were killed in addition to groups III and IV, and 40 percent of the plants in group I had not fully recovered when the final classification was made.

To show that the higher percentage of dead plants in tests 3 and 4 (table 1, C and D) cannot be attributed to the fact that the crosses were made reciprocally, the two types of crosses were tested under identical conditions, rows of one being replicated among rows of the other. Data in both cases gave a good fit to a 1:3 ratio indicating no general difference in their behavior. Results are recorded in table 1, E.

Tests 6 and 7 (table 1, F and G) contain progeny from the backcross (OSF x W-134) x W-134, tested with the highly virulent strain. Test 6 was made in the field in 1934. Approximately one-fourth (22 percent) of the progeny reacted like the susceptible parent (table 1, F). Final results are in close agreement with a 1:3 ratio:

Healthy, 77 (75) : *Dead,* 224 (226), P = .83.

Test 7 was made in the greenhouse. Final results (table 1, G) are also in close agreement with a 1:3 ratio:

Healthy, 195 (187) : *Dead and diseased,* 555 (563), P = .53.

No linkage to the sugary gene could be detected in either of these two tests. The X^2 test for independence applied to the final data showed no significant discrepancies in a four-fold classification, starchy-sugary and healthy-dead. For final data in table 1, F, $X^2 = .18$, P = .68; for final data in table 1, G, $X^2 = 2.48$, P = .12.

Another test of the backcross progeny involving W-134 was made in the field in 1935. Classifications were made on Aug. 19, 48 days after inoculation. At this time the 312 plants of the test were grouped into four classes: Class 1 included plants showing no symptoms; class 2 contained all plants having a high percentage of dried-up leaves but otherwise still green; class 3 comprised those plants on which all leaves were dead, and class 4 consisted of the remaining plants which died early and of which only traces remained. Plants were classified independently of size or vigor. The number of plants in each of the classes was 75, 85, 77 and 75, respectively. This is an excellent fit to

a 1:1:1:1 ratio and again definitely denotes the existence of four major groups of genotypes. One group completely recovers, the other three can be separated on the basis of rate of death. It is, therefore, clearly evident that the same type of segregation occurs in backcross populations involving W-134 as in those involving WF.

REACTION OF F_2 PROGENY FROM CROSSES OSF x WF AND OSF x W-134

F_2 progeny of OSF x WF were tested with the same two strains of bacteria used in the analysis of the backcrosses. Results of inoculations with the strain of low virulence are given in table 2, A. This group of plants was tested at the same time and under the same conditions as the backcross progeny in test 1 (table 1, A). Under conditions giving a 1:1 ratio in the backcross, a 3:1 ratio in the F_2 would be expected. Final results approximate a 3:1 ratio:

Healthy, 432 (447) : *Dead and diseased,* 164 (149), P = .16.

Results obtained with the highly virulent strain under the same environment as the backcross progeny in test 2 (table 1, B), are summarized in table 2, B. Under conditions giving a 1:3 ratio in the backcross, a 9:7 ratio would be expected in the F_2 if two major complementary or supplementary factors were acting. Final results, 35 days after inoculation, exhibit a 9:7 ratio:

Healthy, 266 (252) : *Dead and diseased,* 182 (196), P = .18.

Reactions of F_2 progeny of OSF x W-134 are shown in table 2, C. These were tested at the same time as the backcross progeny in test 7 (table 1, G). It is evident from the number of plants dead after the first 20 days that the disease was quite severe. A few of the remaining plants at the end of the test showed only traces of disease and were classified as doubtful. If the doubtfuls are grouped with the healthy, a 9:7 ratio results:

Healthy, 188 (191) : *Dead,* 152 (149), P = .73.

This 9:7 ratio in the F_2 confirms the backcross ratio of 1:3 (table 1, G) obtained under similar conditions.

REACTION OF OTHER CROSSES

Backcross progeny of (OSF x GB-797) x GB-797 and (Tr x GB-797) x GB-797, and F_2 progeny of Tr x GB-797 were included in the 1934 field tests. Tr was a very resistant, yellow dent inbred in the class with OSF. GB-797 was a susceptible inbred of Golden Bantam sweet corn. Data from these crosses

TABLE 2. RESULTS OF INOCULATIONS OF F_2 PROGENY FROM CROSSES OSF x WF AND OSF x W-134.

Section	Test No. and cross	Virulence of culture	Pedigree number	Endosperm color or character	No. of plants	Early classification			Final classification				
						Days after inoc.	Alive	Dead	Days after inoc.	Heal.	Doubtful	Dis.	Dead
A	(1) OSF x WF F_2	Low	W-819	Yellow	456	22	393	63	42	326		23	107
				White	140	22	121	19	42	106		9	25
				Total	596		514	82		432		32	132
				Percent				13.7		72.4		5.4	22.1
	Parents	Low	OSF	Yellow	34	22	32	2	42	32			2
			WF	White	38	22	18	20	42	1			37
B	(2) OSF x WF F_2	High	W-822	Yellow	332	10	281	51	35	201		14	117
				White	116	10	92	24	35	65		6	45
				Total	448		373	75		266		20	162
				Percent				17.0		59.1		4.4	36.1
	Parents	High	OSF	Yellow	13	10	13	0	35	13			0
			WF	White	9	10	1	8	35	0			9
C	(3) OSF x W-134 F_2	High	W-1159	Starchy	252	20	150	102	40	128	16		108
				Sugary	88	20	47	41	40	43	1		44
				Total	340		197	143		171	17		152
				Percent				42.0		50.3	5.0		44.7

TABLE 3. RESULTS OF INOCULATION OF BACKCROSS AND F_2 PROGENY FROM OTHER CROSSES.

Section	Cross	Pedigree number	No. of plants	Final classification			Proposed ratio	X^2	P
				Heal.	Dis.	Dead			
A	(OSF x GB-797) x GB-797	W-1140	76	27	0	49	3:5	.12	.73
		W-1141	107	28	15	64	2:1:5	.47	.79
							1:3*	.07	.79
B	(Tr x GB-797) x GB-797	W-1120	142	68	0	74	1:1	.25	.63
		W-1121	57	17	0	40	1:3	.70	.42
		W-1122	101	26	0	75	1:3	.03	.87
		W-1123	80	19	0	61	1:3	.06	.80
		W-1124	115	22	0	93	1:3	2.11	.15
		W-1125	190	50	0	140	1:3	.17	.68
		W-1126	128	41	0	87	3:5	1.63	.20
C	Tr x GB-797 F_2	W-1127	177	107	0	70	9:7	1.26	.26
		W-1128	64	41	0	23	9:7	1.58	.21

*Healthy : Dead and diseased.

are summarized in the different sections of table 3. Two ears of the backcross (OSF x GB-797) x GB-797 were tested. In one group of progeny, W-1140, the data (table 3, A) are in agreement with a 3:5 ratio. In the other group, W-1141 (when final classifications were made) a number of the remaining plants showed symptoms of the disease. Division into three classes, healthy, diseased and dead, reveals a 2:1:5 ratio. Apparently conditions were slightly more favorable for disease development in the latter case.

Corresponding ratios were obtained in the backcross (Tr x GB-797) x GB-797. Seven ears of this backcross were tested. Conditions were not the same for each test. The results (table 3, B) show that 1:3 ratios were the most common but in certain tests 1:1 and 3:5 ratios appeared. The 1:3 ratios were verified by the 9:7 ratios in the two groups of F_2 progeny recorded in table 3, C. In the case of W-1128 the observed F_2 ratio of 41:23 will also fit a 45:19 corresponding to a 3:5 ratio in the backcross.

It is apparent that the reaction of the backcross and F_2 populations of these other crosses is similar to that described for backcross and F_2 populations of the crosses OSF x WF and OSF x W-134. This suggests that the major factors responsible for the resistance of Tr or susceptibility of GB-797 do not differ from those concerned in OSF or WF and W-134.

FACTORIAL HYPOTHESIS FOR INHERITANCE OF RESISTANCE

Data from the various backcross and F_2 populations have shown that four major groups of genotypes can readily be differentiated. Under certain conditions, however, it was evident that they were not in themselves a homogeneous lot but could be further divided into two equal groups. Such results may be

explained on the basis of three independently inherited, dominant, supplementary factors. These are represented symbolically in order of their ability to produce resistance as follows:

Sw_1—Major gene for resistance.

Sw_2—When present alone has less effect than Sw_1. In combination with Sw_1 acts supplementarily in production of high resistance.

Sw_3—A minor modifying factor, supplementary or additive in combination with either one or both of the above.

On the basis of this factorial scheme, the genotypes of the parental inbreds probably were:

Resistant—$Sw_1Sw_1Sw_2Sw_2Sw_3Sw_3$

Susceptible—$sw_1sw_1sw_2sw_2sw_3sw_3$

The proposed interaction and phenotypic expression of these factors, in a backcross population, are given in table 4 on the basis that dominance is fairly complete. Genotypes are arranged in a descending scale of resistance from "a" to "h." A major break in this scale of resistance occurs between the genotypes "b" and "c" as shown by 1:3 ratios in the backcross and 9:7 ratios in F_2. A second major break, less sharply differentiated, occurs between genotypes "d" and "e." This is signified by the 1:1 backcross ratios obtained with the strain of low virulence. The 3:1 ratios in F_2 obtained under identical conditions also indicate that "d" is more resistant than "e." A third major break is evident between "f" and "g" if the time of death of

TABLE 4. GENOTYPES PROPOSED FOR BACKCROSS PROGENY AND THEIR PHENOTYPIC EXPRESSION ON THE BASIS OF COMPLETE DOMINANCE.

Group		Proposed genotypes in backcross	Phenotypic expression
I	(a)	$\underline{Sw_1}sw_1\underline{Sw_2}sw_2\underline{Sw_3}sw_3$	Very resistant, fully recovering from infection under ordinary conditions for disease development.
	(b)	$\underline{Sw_1}sw_1\underline{Sw_2}sw_2sw_3sw_3$	
II	(c)	$\underline{Sw_1}sw_1sw_2sw_2\underline{Sw_3}sw_3$	Moderately resistant, tolerate disease for a considerable period but usually die later in development; attacked only by a comparatively virulent strain of the organism under favorable conditions; (d) slightly lower in resistance than (c), shown by 3:5 ratios.
	(d)	$\underline{Sw_1}sw_1sw_2sw_2sw_3sw_3$	
III	(e)	$sw_1sw_1\underline{Sw_2}sw_2\underline{Sw_3}sw_3$	Susceptible under a wider range of conditions; attacked by less virulent strains of the organism differentiating it from above; (f) slightly lower in resistance than (e).
	(f)	$sw_1sw_1\underline{Sw_2}sw_2sw_3sw_3$	
IV	(g)	$sw_1sw_1sw_2sw_2\underline{Sw_3}sw_3$	Very susceptible, dying shortly after inoculation; reaction corresponds to that of the susceptible parent.
	(h)	$sw_1sw_1sw_2sw_2sw_3sw_3$	

the susceptible parent is taken as a basis. Apparently the genotype containing only the dominant Sw_3 is but slightly more resistant than the triple recessive.

VERIFICATION OF FACTORIAL HYPOTHESIS BY TESTS OF BACKCROSS AND F_3 FAMILIES

Three hundred and eight families from the backcross (OSF x W-134) x W-134, 97 families from the backcross (OSF x WF) x WF, 196 F_3 families from the F_2 of OSF x W-134 and 84 F_3 families from the F_2 of OSF x WF were tested for resistance. Most of the tests were made in the greenhouse. Generally, from 48-52 families (40-50 plants per family) were tested on one of the large benches at the same time. Two such benches were used. Since the environment was not the same for both benches or for tests made at different times, each group of 48-52 families was analyzed separately. Unless otherwise stated, each test involved a different group of families.

Frequency distributions based on percentage of dead plants per family (5 percent classes) are presented in the form of histograms. Two distributions are usually given for each test. One shows the early distribution at a time when the majority of the plants of the susceptible check were dead. The other shows the final distribution of the same families at a later date when wilt had ceased.

Reaction of the parental inbreds is shown in each histogram by means of little squares above the classes in which they fell. The black portion of each bar represents the proportion of families from red-cobbed plants in each class. If there was no association between red cob and resistance, the ratio of families from red-cobbed plants to those from white-cobbed plants in each class, theoretically, should be 1:1 in tests of backcross families and 3:1 in tests of F_3 families. This part of the data will be discussed later under linkage relations.

BREEDING BEHAVIOR OF BACKCROSS FAMILIES

Early distributions of the backcross families in the various tests are given in fig. 6, A, B, C, D and E, and fig. 7, F, G, H and I. Final distributions, when wilt had ceased, are given in A_1, B_1, C_1, D_1 and E_1 of fig. 6, and F_1, G_1, H_1 and $I_1$of fig. 7, respectively. Each of the early and final distributions is discussed separately. The points taken as breaks in distribution are not mathematically significant, but, as will be pointed out later, trends are similar in all tests. Final conclusions are based on the bulk trend.

Tests 1 and 2 were run at the same time but on separate benches. Families of test 1 were replicated in test 2. In the

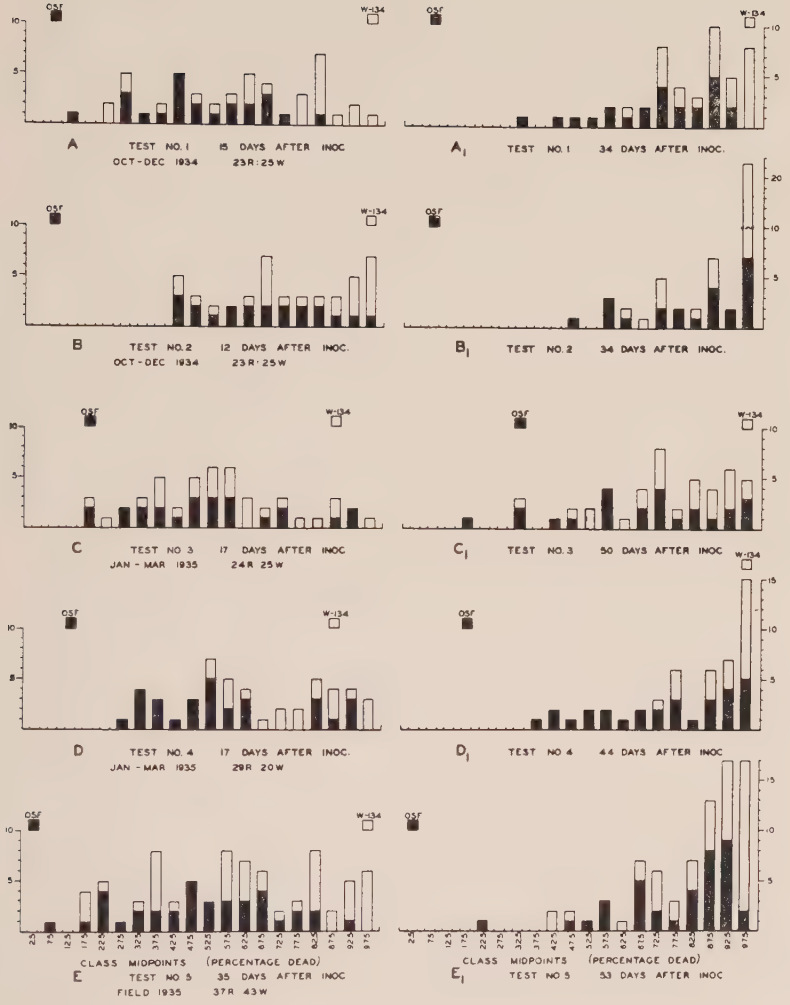

Fig. 6. Histograms showing the distribution of backcross families with respect to percentage of dead plants per family in tests Nos. 1-5. (R = families from red-cobbed plants; W = families from white-cobbed plants. The ratio of R to W in each class is shown by the ratio of black to white in each of the bars of the histograms.)

early distribution of test 1 (fig. 6, A) four modes are evident, indicating four major types of segregation with about one-fourth of the families in each category. The early distribution of test 2 (fig. 6, B) is not exactly comparable to this, but the same trends are indicated. First counts were made in both tests on the same day. In test 2, however, plants died much more rapidly, and more plants were dead when the first classifications were made.

Tests 3 and 4 constitute the second plantings on the two benches. In the early distribution of test 3 (fig. 6, C) 24 of the 49 families in the test fell within the class range from 42.5 to 78.5 percent dead, points which mark slight breaks in the distribution. Eleven of the remaining families are above the upper range and 14 below the lower range of this group. This distribution may be explained if one assumes that only plants of the very susceptible genotypes "g" and "h" (table 4) within the various families were dead when classifications were made. In this case the backcross families, representative of group I (table 4), would segregate on a 15:1 basis, those of groups II and III on a 3:1 basis, and those of group IV would all be dead if only the proposed major factors were involved. Thus the 49 families should fall into three groups in the proportion 12:25:12. The actual distribution was 11:24:14.

In test 4, although a greater number of plants were dead when the first classifications were made, two slight breaks in distribution (fig. 6, D) are again visible. The first break from left to right includes eight families. The second divides the remaining equally. Such a distribution may be explained by assuming that plants of the susceptible genotypes "f," "g" and "h" among these families were dead when first counts were made. On this basis, the backcross plants of genotype "a" upon selfing should segregate 57:7; those of genotypes "b," "c" and "d" should segregate 3:1; those of genotype "e" should segregate 9:7, and those of genotypes "f," "g" and "h" should all die. If the first break separates the families segregating 57:7 from those segregating 3:1 or 9:7 and the second break separates the group segregating 3:1 or 9:7 from those that should all die, the distribution would be in accordance with a 1:4:3 ratio. Theoretically, therefore, the 49 families within the test should be in the ratio of 6:25:18. Actually, the ratio was 8:21:20 ($X^2 = 1.52$, $P = .45$).

Test 5 was made in the field. Sixty-two of the eighty families within the test had been tested previously in the greenhouse. Several breaks in the early distribution (fig. 6, E) may be distinguished. Approximately one-fourth (22) of the families fell within the range from 7.5 to the second slight break at the 42.5 percent point. The first break at the 27.5 percent point divides

this group into two equal parts. On the other end of the scale approximately another fourth fell within the range from the break at the 72.5 percent point to 100. This group is also divided almost equally by a break at the 87.5 percent point. Such a distribution may be interpreted as showing breaks in segregation between families from plants of genotypes "a" and "b," "b" and "c," "f" and "g," and "g" and "h" (table 4) resulting in a 1:1:4:1:1 ratio. The actual distribution as separated by the breaks was 11:11:34:11:13, in agreement with expectations ($X^2 = 2.1$, $P = .70$). In this case there is direct evidence for the existence of at least eight different genotypes or groups of closely related genotypes in a backcross population.

In tests 6 and 7 the disease was very mild. The early distributions (fig. 7, F and G) are not comparable to those of other tests. It may be noted that the final distributions (fig. 7, F_1 and G_1) are similar to what one would expect in the early distributions. Apparently only plants of very susceptible genotypes died throughout the course of these two tests.

Tests 8 and 9 include families from the backcross (OSF x WF) x WF. Test 9 is a replication of test 8. It is obvious (fig. 7, H and I) that the early distributions of these families are similar to those of the backcross families involving W-134. In fig. 7, I, four modes may be distinguished indicating four major types of segregation.

Further verification of the proposed genotypes or groups of genotypes of table 4 is manifest in the final classification of families for each of the tests, except 6 and 7. The first break in the final distributions from the right end of the scale usually divides the families into approximately two equal groups. This may be interpreted as a break in segregation between families from plants representative of groups II and III. The break between families from plants representative of groups III and IV is generally obliterated because those from group III, as they approach 100 percent dead, overlap those from group IV.

In test 1 (fig. 6, A_1), if one disregards the slight drop in frequency in the 92.5 percent class, the first break from the right comes at the 82.5 percent class. Fifty-four percent of the families fell within the range 100 to 81 inclusive (reading from high to low percentages dead per family). If the slight drop in the 92.5 percent class is indicative of a break, however, families from plants of groups III and IV may still be differentiated. Twenty-seven percent of the families fell within the range 100 to 91 and 27 percent within the range 90 to 81.

In test 2 (fig. 6, B_1), 48 percent of the families had died out completely when the final classification was made. In test 3

(fig. 6, C_1), the first low point in distribution from the right comes at the 77.5 percent class. The range from 100 to 76 includes 46 percent of the families. In test 4 (fig. 6, D_1), the first break occurs at the 82.5 percent class. The range from 100 to 86 includes 57 percent of the families.

Breaks at similar points are apparent in final distributions of families involving WF, tests 8, 9 and 10. In test 8 (fig. 7, H_1), the first break from right to left again comes at the 77.5 percent class. The range from 100 to 81 includes 59 percent of the families. In test 9 (fig. 7, I_1), the first low point from right to left also comes at the 77.5 percent point, but approximately three-fourths of the families fell within the range up to this point. There is, however, a very sudden drop in frequency after the 92.5 percent class. The range from 100 to 91 includes 53 percent of the families. In test 10 (fig. 7, J_1), the first break from right to left comes at the 82.5 percent class. Fifty-four percent of the families fell within the range to this point. Thus it may be seen that approximately half of the families in most of the tests are included within the range up to the first break in distribution from the right end of the scale even though these breaks may come at different points.

Families from selfed plants of group II (table 4) may be distinguished in a similar manner. In most of the final distributions approximately one-fourth of the families fell between the first and second breaks from right to left. For example, in fig. 6, A_1, 25 percent of the families fell within the range 80 to 71; in fig. 6, B_1, 23 percent were within the range 95 to 81; in fig. 6, C_1, 27 percent fell within the range 75 to 61; in fig. 6, D_1, 25 percent were within the range 85 to 66, and in H_1, I_1 and J_1 of fig. 7, 22, 22 and 24 percent, respectively, were in the range between the first and second breaks from the right. In the final distribution of test 5 (field test, fig. 6, E_1), the first break in distribution from right to left comes at the 77.5 percent class. The range up to this point includes 72 percent of the families. In this case no break appeared at a point dividing families representative of group II from those of groups III and IV.

Families representative of group I may be distinguished as the remaining families above the second break in distribution from the right except in test 5. In test 5 (fig. 6, E_1), they may be distinguished as those above the first break, since this came at a point which separated a group including approximately three-fourths (72 percent) of the families within the test. In fig. 6, A_1, the remaining 21 percent are scattered over a wide range from 70 to 31. In fig. 6, B_1, the remaining 29 percent seem to be divided into two equal groups, one group with a mode at about 72 and the other with a mode around 57. This suggests two types of segregation in group I. The group with a mode

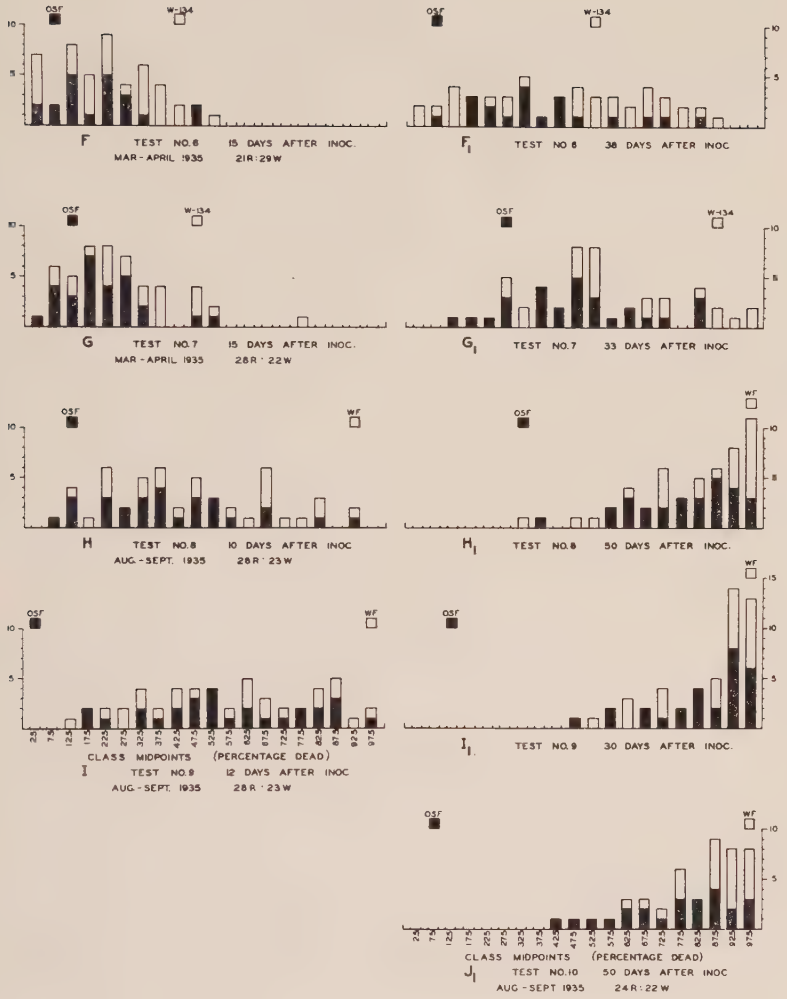

Fig. 7. Histograms showing the distribution of backcross families with respect to percentage of dead plants per family in tests Nos. 6-10.

around 57 can be fitted to a 27:37 ratio indicating that at least one-eighth of the backcross plants had genotypes similar to the F_1 plants in accordance with the interaction of three dominant supplementary factors. Similar trends indicating two types of segregation in group I are evident in C_1, D_1 and E_1 of fig. 6 and H_1 and J_1 of fig. 7. In fig. 6, C_1, the remaining 27 percent above the second break are scattered over the range from 60 to 16 with modes at 55 and 30. In fig. 6, E_1, 28 percent of the families fell within the range 75 to 21 with a break at the 62.5 percent class.

The numbers of families within the groups separated by the first and second breaks from the right end of the scale in the final distribution of each test except 5, 6 and 7 are summarized in table 5. Assuming that the first break from the right comes between families representative of groups II and III of table 4 (break between III and IV not shown because of over-lapping) and that the second break comes between families representative of groups I and II, the theoretical distribution should be in accordance with a 1:1:2 ratio. Numbers enclosed in parentheses in the table give the frequencies expected on the basis of this ratio.

Consistency of the 1:1:2 ratios in table 5 signifies that at least four major types of segregation occur in a representative group of families from selfed backcross plants. That each of these may be further subdivided into two minor groups is shown in several of the early classifications (fig. 6, D and E) and by the break in distribution of families supposedly representative of group I, as shown in certain of the final classifications. The

TABLE 5. CLASSIFICATION OF FAMILIES INTO GROUPS ACCORDING TO BREAKS IN FINAL DISTRIBUTIONS OF THE VARIOUS TESTS.

Test No.	Number of progenies	Major segregating groups			X^2	P
		I	II	III-IV		
1	48	10 (12)	12 (12)	26 (24)	0.49	0.78
2	48	14 (12)	11 (12)	23 (24)	.45	.80
3	48	13 (12)	13 (12)	22 (24)	.34	.85
4	49	9 (12)	12 (12)	28 (25)	1.11	.58
8	51	10 (13)	11 (13)	30 (26)	1.60	.45
9	51	13 (13)	11 (13)	27 (26)	.33	.85
10	46	10 (11)	11 (11)	25 (23)	.26	.87
Total		79 (85)	81 (85)	181 (171)	1.18	.56

breaks in distribution taken as division points are very slight and probably not significant if only one test is considered. But, inasmuch as the number of plants was small, and since the breaks repeatedly came at definite points, dividing the families into similar groups in each case, they certainly may be regarded as significant. Furthermore, groups I and II were shifted to the right and the breaks narrowed because of death due to agents other than *Phyt. stewarti*. Invariably a number of seedlings per family were killed by other pathogens such as *Diplodia zeae* (Schw.) Lev., *Gibberella saubinetii* (Mont.) Sacc., *Fusarium moniliforme* (Sheld.), or a species of *Penicillium* thought to be *Penicillium oxalicum* Currie and Thom. These pathogens could be partially controlled in the resistant check, OSF, by careful selection of seed for planting, but in the case of F_3 or backcross families this could not be done because of the limited supply of seed per family. Even with careful selection of seed in the resistant check, it is evident from the various distributions that a number of plants were killed. One may conclude, therefore, that the breeding behavior of the backcross families supports the factorial hypothesis proposed in table 4 insofar as the interactions of the three major factors, Sw_1, Sw_2 and Sw_3, are concerned. It is apparent, however, that other minor modifying factors may also be acting.

BREEDING BEHAVIOR OF F_3 FAMILIES

Frequency distributions of various tests of F_3 families are given in figs. 8 and 9 (tests 12-18). Four distributions of test 12 are shown (fig. 8, A-A_3). Figure 8, A, shows the early distribution when the majority of the plants of the susceptible parent W-134 were dead. At this time the majority of the plants within 2 of the 51 families were also dead. This suggests that a few F_2 plants are similar in genotype to that of the susceptible parent. On the basis of three independent factors, the triple recessive theoretically would be expected to occur as frequently as 1 in 64. According to previous data, the genotypes of the combination $sw_1sw_2Sw_3$ offer little resistance. Consequently, 1 out of every 16 families would be expected to die out completely at an early date. Therefore, one might expect 3 of the 51 families to react like the susceptible check, whereas two were observed.

Figure 8, A_1, shows the reaction of the same test 22 days after inoculation. A gradual shift to the right in distribution can be seen. Figure 8, A_2, shows the distribution 45 days after inoculation. Here a few of the families remain similar in reaction to the resistant parent. From this point on, the distribution curve rises in somewhat irregular fashion, reaching the first peak at

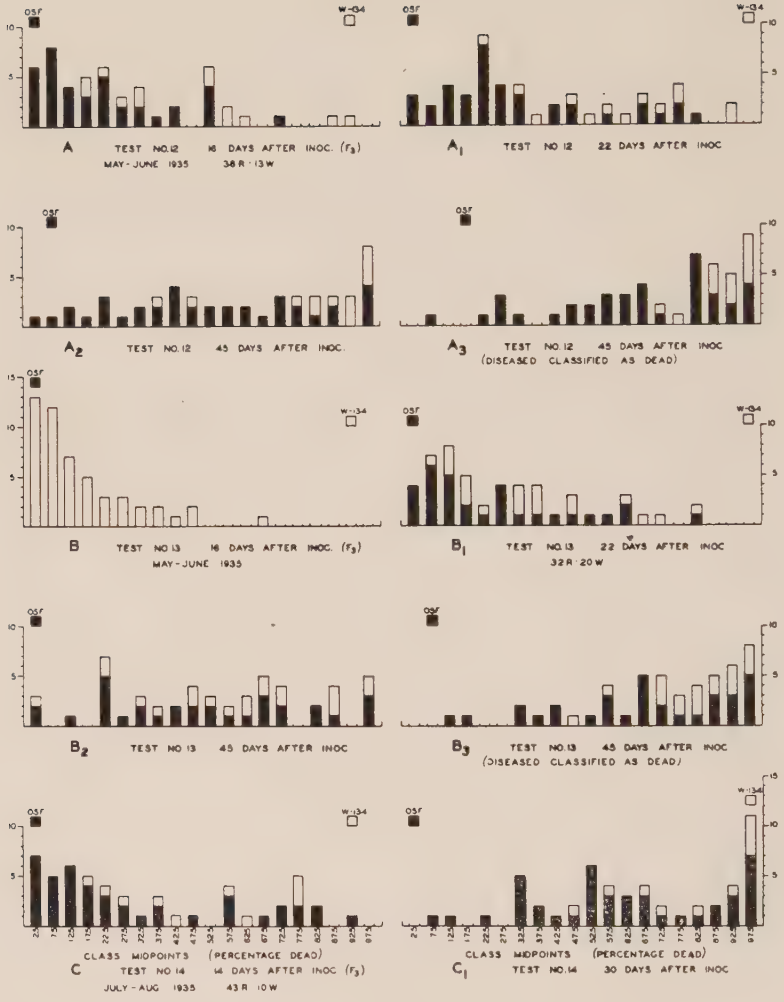

Fig. 8. Histograms showing the distribution of F_3 families with respect to percentage of dead plants per family in tests Nos. 12-14.

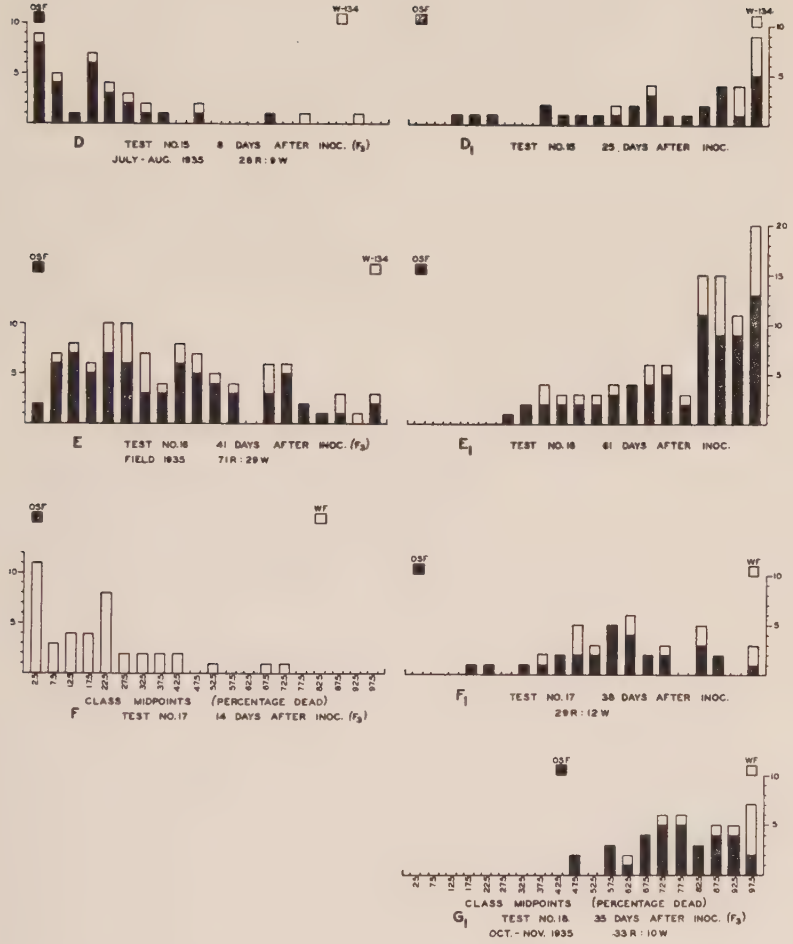

Fig. 9. Histograms showing the distribution of F_3 families with respect to percentage of dead plants per family in tests Nos. 15–18.

the 42.5 percent class, then gradually falling off to another low at the 67.5 percent class before again ascending. If the low point at the 67.5 percent class may be taken as an indication of a major break in type of segregation, results are in agreement with a 9:7 ratio:

Families with 1-70 percent dead per family 28 (29) :
Families with 71-100 percent dead per family 23 (22).

Figure 8, A_3, shows the final distribution in which diseased plants were classified as dead. Here again it is apparent that certain families react like the resistant parent. Disregarding the break at the 37.5 percent class, the distribution curve gradually ascends to the 67.5 percent class, then declines to a low at 77.5 and again rapidly rises. If the low point at the 77.5 percent class (a point similar to that taken in A_2) is interpreted as a major break in segregation, the distribution approaches a 27:37 ratio:

6-80 percent dead 24 (22) : *81-100 percent dead* 27 (29), P = .48.

Four distributions for test 13 also are given (fig. 8, B-B_3). The disease was not so severe in this test, but the same general trends are evident. It may be noted that the point taken as indicative of a break in segregation in A_2 and A_3 of test 12 was the first low point in distribution from the right end of the scale. A similar point in the final distribution of test 13 (fig. 8, B_3) again divides the families in accordance with a 9:7 ratio:

11-80 percent dead 27 (28) : *81-100 percent dead* 23 (22).

Tests 14 and 15 (C and C_1 of fig. 8, and D and D_1 of fig. 9) were made during July and August, 1935. Temperatures were high and susceptible plants wilted quickly, but the same trends appeared. Final distributions of test 14 are shown in fig. 8, C_1. By again taking the first low point in distribution from right to left as a break in segregation, the results are in agreement with a 9:7 ratio:

5-80 percent dead 32 (29) : *81-100 percent dead* 20 (23), P = .45.

In test 15 the division point in the final distribution (fig. 9, D_1) comes at nearly the same place in the scale. Seventeen of the thirty-seven families in the test are within the range 10-75 and 20 within the range 76-100 percent dead. This is an excellent fit to a 27:37 ratio.

Final distribution of the 100 F_3 families tested in the field is given in fig. 9, E_1. Here again a slight break is evident at the

77.5 percent class. Thirty-nine families fell within the range 25-80 and 61 within the range 81-100 percent dead. This distribution likewise approaches a 27:37 ratio ($X^2 = .41$, $P = .52$). In this field test one would expect a few of the families to remain within the range 1-25 percent dead. Final notes were taken Sept. 10. At this time it was rather difficult to distinguish between plants showing dead leaves due to maturity and those showing dead leaves due to the pathogen. Classifications made 2 weeks previously denoted a very low percentage dead in many of the families. In the final classification, therefore, many of the plants may have been misclassified because of maturity, since the families were segregating for maturity as well as resistance.

Tests 17 and 18 include F_3 families from the F_2 of OSF x WF In test 17 the disease was not very severe. It seems as if there might be a slight break in the final distribution (fig. 9, F_1) at the 67.5 percent class. If this point may be taken as a break in type of segregation, the distribution is in agreement with a 45:19 ratio ($X^2 = .9$, $P = .31$).

In test 18 (fig. 9, G_1), the disease was much more severe. Trends similar to tests of F_3 families involving W-134 are noticeable. The first break in distribution from right to left divides the families into a 9:7 ratio:

45-80 percent dead 23 (24) : *81-100 percent dead* 20 (19).

In this test and in many of the other greenhouse tests of F_3 families, it is evident that certain families as well as the resistant check, OSF, were shifted much farther to the right on the scale than one would expect. This may be attributed to death caused by agents other than *Phyt. stewarti.*

In general, the results obtained in various tests of F_3 families are also in keeping with the hypothesis based on the interaction of three dominant factors. Certain families, as frequent as 1 in 64, behave like the resistant parent or breed true for resistance (fig. 10, A). Others, also as frequent as 1 in 64, behave like the susceptible parent breeding true for susceptibility (fig. 10, B). Between these two extremes various types of segregation are evident. Figure 11 shows a segregating family in which the majority of the plants are susceptible. Under severe conditions a break in type of segregation differentiates the families on the basis of a 27:37 ratio. The division apparently comes between families from F_2 plants containing all three dominant factors, and those from plants containing only two dominant factors or less. Under less severe conditions as shown by the 9:7 ratios the division apparently comes between families from F_2 plants containing either all three factors or the two Sw_1Sw_2,

and those containing only Sw_1 or Sw_2, or Sw_1 and Sw_2 in combination with Sw_3.

LINKAGE RELATIONS

COB COLOR AND RESISTANCE

Relations between cob color ($P^{wr}p$) and resistance were determined entirely from tests of backcross and F_3 families. The remaining plants in a field test of backcross or F_2 populations would afford some data on cob color relations, but numbers in these tests were usually small and such data were not recorded.

Theoretically, half of the backcross families should segregate 3 red : 1 white, and the other half should breed true white with respect to cob color. If there was no association between cob color and resistance, the two types of families should be equally distributed throughout the various classes of the frequency distributions shown in figs. 6 and 7. The proportion of families from red-cobbed plants to those from white-cobbed plants in any one class is shown by the ratio of black to white, respectively, in each bar of the histograms.

It may be seen that more black appears on the left end of the scale than on the right, showing that families from red-cobbed plants tend to be congregated on the resistant end, whereas

Fig. 10. A (top). An F_3 family breeding true for resistance. B (bottom). An F_3 family breeding true for susceptibility. (Photographed 5 weeks after inoculation.)

Fig. 11. An F_3 family segregating for resistance and susceptibility. (Photographed 5 weeks after inoculation.)

those from white-cobbed plants tend to be assembled on the susceptible end of the scale. A quantitative estimate of this relation may be obtained from table 6 where the data from the early classifications of tests 1, 3 and 4 (fig. 6, A, C and D) and the two comparable final classifications of tests 6 and 7 (fig. 7, F_1 and G_1) are lumped into 10 percent classes. The value of P reveals a highly significant difference between the distribution of the two types of families. A similar highly significant difference is shown in table 7 where the data from the final classifications of tests 2, 3, 4 and 5 (fig. 6, B_1, C_1, D_1 and E_1) are lumped into 10 percent classes. Final data of test 1 were not included because the same families were represented in test 2.

TABLE 6. COMPARATIVE DISTRIBUTIONS OF FAMILIES FROM SELFED RED AND WHITE-COBBED PLANTS OF THE BACKCROSS INVOLVING W-134.

(Early classifications.)

Class midpoints (Percentage dead)	5	15	25	35	45	55	65	75	85	95	Total
Seg. 3R : 1W	1	9	13	22	25	20	13	5	10	5	123
Breeding true W	3	4	10	8	11	24	17	15	20	11	123
Total	4	13	23	30	36	44	30	20	30	16	246

$X^2 = 26.8$. P = less than .01.

TABLE 7. COMPARATIVE DISTRIBUTIONS OF FAMILIES FROM SELFED RED AND WHITE-COBBED PLANTS OF THE BACKCROSS INVOLVING W-134.

(Final classifications.)

Class midpoints (Percentage dead)	5	15	25	35	45	55	65	75	85	95	Total
Seg. 3R : 1W		1	1	3	7	15	11	17	24	34	113
Breeding true W				1	4	2	8	18	22	58	113
Total		1	1	4	11	17	19	35	46	92	226

$X^2 = 20.2$, N = 6 (first four classes were lumped)
P = less than .01.

Significant differences in distribution of the two types of families are also exhibited in tests of backcross families involving WF (fig. 7, tests 8, 9 and 10). Final data of tests 8 and 10 (fig. 7, H_1 and J_1) combined into 10 percent classes are shown in table 8. Test 9 was a duplicate of test 8 and, therefore, was not included.

TABLE 8. COMPARATIVE DISTRIBUTIONS OF FAMILIES FROM SELFED RED-COBBED AND WHITE-COBBED PLANTS OF THE BACKCROSS INVOLVING WF.

(Final classifications.)

Class midpoints (Percentage dead)	5	15	25	35	45	55	65	75	85	95	Total
Seg. 3R : 1W				1	2	4	9	9	15	11	51
Breeding true W				1	1	1	3	8	8	24	46
Total				2	3	5	12	17	23	35	97

$X^2 = 11.6$, N = 4 (classes 35, 45, 55 were lumped)
P = .02

In the case of F_3 families, if resistance and cob color were independent, families from white-cobbed plants should be distributed throughout each of the various classes of a frequency distribution in the proportion of 1 to 3. It may be seen in the various histograms of figs. 8 and 9 that the majority of the families from white-cobbed plants are on the susceptible end of the scale.

An association between red cob color and resistance, therefore, is definitely evident in both backcross and F_3 families. Since certain red-cobbed families breed true for complete susceptibility, there is no reason to suppose any great physiological interplay between cob color and resistance. The cause of the

association, then, may be due to genetic linkage. A critical test would be to analyze a cross in which resistance was brought in with white cob.

SUGARY ENDOSPERM AND RESISTANCE

Separate analyses of plants from starchy and sugary seeds in the backcross and F_2 generations (tables 1 and 2) showed no indications of association between susceptibility and the sugary gene. The analyses of families of selfed, backcross plants confirm these results. As in the cob color relations, 50 percent of the families of selfed backcross plants should segregate on the basis of three starchy (Su) to one sugary (su) and the other 50 percent should breed true for sugary. Both of these types ought to be uniformly distributed over the scale of resistance if endosperm character is independent of resistance or susceptibility.

Table 9 shows the comparative distributions of the above two family types among the same group of families represented in table 6. Families are much more uniformly distributed in regard to endosperm character than cob color. There is, however, a slight tendency in these early classifications for a few more of the families breeding true for sugary to be congregated on the susceptible end of the scale than there is in the case of those segregating 3 Su : 1 su. Statistically, the differences are not significant.

At the time of final classifications (table 10), the two types of families are distributed almost equally. Here, there is no indication whatsoever of linkage between endosperm character and resistance. Since the seedlings from sugary seeds were usually weaker at the time of inoculation than those from starchy seeds, a more rapid rate of death would be expected in families breeding true sugary. This probably accounts for the slight association in the early classifications.

TABLE 9. COMPARATIVE DISTRIBUTIONS OF BACKCROSS FAMILIES SEGREGATING 3Su : 1su AND THOSE BREEDING TRUE su.

(Early classifications.)

Class midpoints (Percentage dead)	5	15	25	35	45	55	65	75	85	95	Total
Seg. 3Su : 1su	3	10	15	16	12	26	14	8	13	6	123
Breeding true su	1	3	8	14	23	18	16	12	17	10	122
Total	4	13	23	30	35	44	30	20	30	16	245

$X^2 = 14.4$, N = 9, P = .10

TABLE 10. COMPARATIVE DISTRIBUTIONS OF BACKCROSS FAMILIES SEGREGATING 3Su : 1su AND THOSE BREEDING TRUE su.

(Final classifications.)

Class midpoints (Percentage dead)	5–35	45	55	65	75	85	95	Total
Seg. 3Su : 1su	3	4	9	10	17	25	46	114
Breeding true su	3	7	8	9	18	21	46	112
Total	6	11	17	19	35	46	92	226

$X^2 = 1.31$, $P = .97$.

DAYS TO MATURITY AND RESISTANCE

It has been pointed out in the review of literature and in the results of inbred tests that most of the early varieties and early yellow inbreds of sweet corn are susceptible. This suggests an association between early maturity and susceptibility. In the 1935 field tests, relative maturity of 78 families from selfed plants of the backcross (OSF x W-134) x W-134 was determined on the basis of number of days to silking. Families were divided into three groups as follows:

Group	Number in each group	Percent of total
Early, silking within 57 to 59 days	21	27
Medium, silking within 60 to 62 days	33	42
Late, silking within 63 to 68 days	24	31
	78	100

Distribution of each of the groups with respect to resistance is given separately in fig. 12 together with the theoretical distribution on the basis of complete independence. Theoretical distribution of each group was calculated from the final distribution of the 78 families in 10 percent classes on the basis that the total frequencies in any one class consist of early, medium and late families in the ratio of 27:42:31. An association between resistance and maturity is obvious. The early maturing families (fig. 12, A) tend to be assembled on the susceptible end, whereas the late families (fig. 12, C) tend to be assembled on the resistant end of the scale. The distribution of the intermediate families (fig. 12, B) coincides fairly well with its theoretical value.

EAR ROW NUMBER AND RESISTANCE

Lindstrom (4) has presented evidence that a gene for number of rows very likely resides on chromosome I reasonably near the P gene for red cob. Since it has been shown that one of the major genes for resistance may also be on chromosome I, an association between row number and resistance might be expected. Data in fig. 13, however, do not show a marked association between resistance and row number, although some relation is apparent. The backcross families from 8-rowed plants (fig. 13, A) are in slight excess on the susceptible end of the scale, while those from 12-rowed plants (fig. 13, C) slightly favor the resistant half of the scale. No definite trend is shown by families from 10-rowed plants (fig. 13, B).

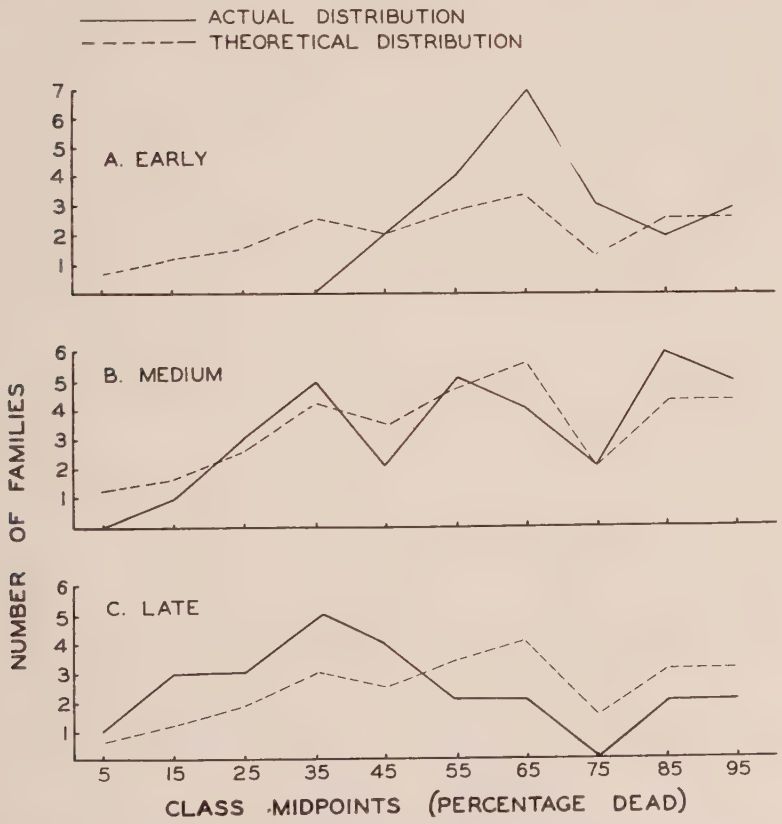

Fig. 12. Frequency distributions of early, medium and late maturing backcross families.

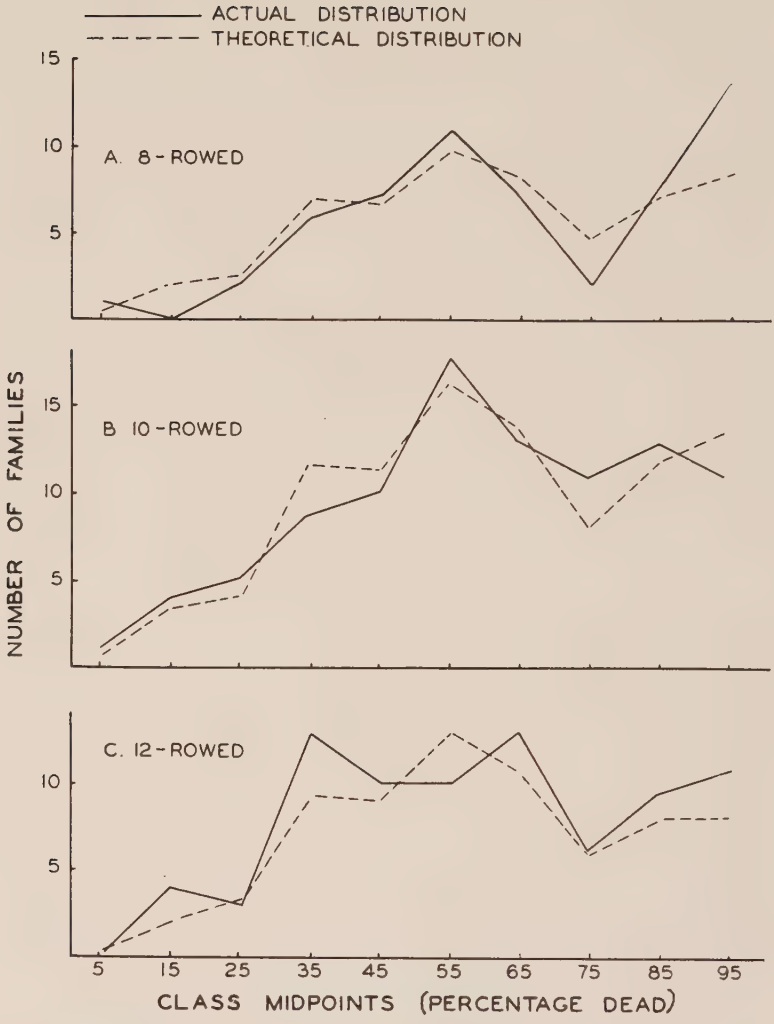

Fig. 13. Frequency distributions of 8, 10 and 12-rowed backcross families.

Distributions in fig. 13 are based on the data in fig. 6, B, C, D and E combined into 10 percent classes. Among the 228 families thus combined, 58 were from 8-rowed, 95 from 10-rowed and 75 from 12-rowed plants. Theoretical distributions were calculated on the basis that the three types were represented in each class in the proportion 58:95:75.

DISCUSSION AND CONCLUSIONS

Data in the foregoing pages clearly show that at least three independently inherited dominant supplementary factors are involved in the inheritance of resistance to bacterial wilt in the two crosses OSF (very resistant dent) x WF (very susceptible flint) and OSF x W-134 (very susceptible sweet). The three factors were designated Sw_1, Sw_2 and Sw_3 in the order of importance. The symbol Sw was derived from the more specific name of the disease "Stewart's wilt."

The hypothesis as proposed in table 4 seems to be justified insofar as the action and interaction of the three factors are concerned. When all three factors are present in the dominant condition a high degree of resistance results, such as that shown by the resistant inbred OSF. When none of the dominant factors is present a high degree of susceptibility results, such as that shown by the two susceptible inbreds WF and W-134. Dominance is fairly complete. The heterozygote $Sw_1sw_1Sw_2sw_2Sw_3sw_3$ appears to be fully as resistant as the homozygote $Sw_1Sw_1Sw_2Sw_2Sw_3Sw_3$.

Intermediate degrees of resistance that are manifest in the F_2 and backcross populations may be explained by the presence of only one dominant factor and by the different possible combinations of any two. Apparently each factor when present alone is capable of producing a certain degree of resistance. In this respect, however, the three factors are not equal. Genotypes containing Sw_1 alone, either in heterozygous or homozygous dominant condition, are more resistant than those containing Sw_2 alone. Genotypes containing only Sw_2 are more resistant than those containing Sw_3 alone. Genotypes possessing Sw_3 by itself are only slightly more resistant than the triple recessive types.

These factors, when in combination, seem to act supplementarily or additively. Sw_1 combined with Sw_2 results in a much higher degree of resistance than when either is present singly. The addition of the third factor Sw_3 increases the resistance still further although only slightly. Genotypes with the combination Sw_1Sw_2 can be distinguished from those with the combination $Sw_1Sw_2Sw_3$ only with a highly virulent culture under very favorable conditions for disease development. Sw_3 combined with Sw_1 or Sw_2 also results in a slightly higher degree of resistance

than when either factor is present alone. In other words, the combination Sw_1Sw_3 brings about a higher degree of resistance than Sw_1 alone, but not as high as that resulting from the combination Sw_1Sw_2. Similarly, the combination Sw_2Sw_3 produces higher resistance than Sw_2 alone but is probably not capable of producing a degree of resistance as high as that produced by Sw_1 alone.

The fact that other crosses such as Tr x GB-797 revealed a similar type of segregation would indicate that the basic mechanism for inheritance of resistance as outlined above is general. The source of the inbreds Tr and GB-797 is widely different from that of OSF and WF.

In a preliminary paper (12) it was shown that the 56 different inbred lines tested may be divided into five classes ranging from highly resistant to highly susceptible. If the three-factor hypothesis can be applied to inbreds in general, the five classes of resistance may be explained genetically as follows: The highly resistant inbreds of class 1, such as OSF and Tr, perhaps contain all three dominant supplementary factors Sw_1, Sw_2 and Sw_3. Inbreds of class 2, slightly less resistant, may lack the minor factor Sw_3. The intermediate class 3 probably consists chiefly of inbreds possessing the combination Sw_1Sw_3 or Sw_2Sw_3, and those having Sw_1 alone. The susceptibile class 4 probably contains the inbreds with Sw_2 or Sw_3 alone, and class 5 probably consists largely of the very susceptible triple recessive types. The genetic situation probably is not as simple as this, for it is quite certain that other minor modifying factors are also involved.

If the above hypothesis is correct, an increase in resistance in crosses of certain intermediate lines would be expected. Unfortunately, only small numbers of such crosses were made and tested. A few of these, however, showed greater resistance than either parent which may be attributed to supplementary or additive action of resistance factors. Although hybrid vigor or heterosis has been emphasized by Mahoney and Muncie (5) as a probable factor in resistance, it does not seem feasible to attribute the increase in resistance of these crosses to hybrid vigor, especially since crosses of other intermediate inbreds were equally vigorous but exhibited no marked increase in resistance. Furthermore, crosses of very susceptible inbreds usually were much more vigorous than their respective parents but showed relatively no increase in resistance. Thus it seems that vigor is not an important factor in determining resistance.

Ivanoff and Riker (2) have presented data showing that, as a rule, the tall, late maturing strains of maize are more resistant than short, early maturing strains. The correlation, however, was not complete for a few relatively short, early maturing

strains were found to have a high degree of resistance and a few tall, late maturing strains had a low degree of resistance. The author (12) also found early resistant strains and late susceptible strains among the 56 inbred lines tested previous to this inheritance study. Since tall, late, susceptible strains and short, early, highly resistant strains exist, one might conclude that genes for maturity and height cannot have much effect on resistance.

In both of the crosses analyzed in this investigation, late maturity was brought into the cross with resistance and early maturity with susceptibility. Tests of backcross families have shown that these characters do not segregate independently. Whether this may be entirely attributed to genetic linkage of factors for maturity with factors for resistance, or whether certain of the factors for maturity may also influence resistance is not definitely known at this stage of the investigation. A critical test would be to analyze crosses in which late maturity was brought in with susceptibility and early maturity with resistance. These crosses have been made but as yet have not been tested. However, as shown in fig. 12, C, a few of the late maturing backcross families were very susceptible, definitely indicating that factors responsible for late maturity cannot have much effect on resistance. The situation seems to be different with factors for early maturity. It is evident from fig. 12, A, that certain of the early maturing backcross families were more resistant than others, but none was as resistant as some of the late maturing families. This suggests that early maturity has some effect.

It has been demonstrated that the parental combinations, red cob and resistance or white cob and susceptibility, were more frequent in the F_2 and backcross populations than the non-parental. The factors for red or white cob cannot have any direct effect on resistance or susceptibility because some of the red-cobbed F_2 or backcross progeny were as susceptible as the most susceptible white-cobbed plants, and *vice versa,* certain white-cobbed plants were as resistant as the most resistant red-cobbed plants. It is highly probable, therefore, that the association is due to genetic linkage and that one of the major factors for resistance is on chromosome I with the P gene for cob color. The linkage cannot be very close for the cross-over types are fairly frequent.

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